## SEQUENCE LISTING

1

| <110>                     | HORIUCHI, TAKASHI<br>KOBAYASHI, TAKEHIKO          |    |
|---------------------------|---|----|
| <120>                     | METHOD FOR AMPLIFYING FOREIGN GENES               |    |
| <130>                     | 084335/0135                                       |    |
|                           | 09/807,409<br>2001-04-13                          |    |
|                           | JP 10/292697<br>1998-10-15                        |    |
|                           | PCT/JP99/05673<br>1999-10-14                      |    |
| <160>                     | 28  |    |
| <170>                     | PatentIn Ver. 2.1                                 |    |
| <210><211><212><212><213> | 28  |    |
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| <400>                     | L<br>gacgt tgcggccata tctaccag                    | 28 |
| 9-9                       | ,yy-gg  |    |
| <210><211><212><213>      | 40  |    |
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| <210><211><212><213>      | 40  |    |
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| agatt          | gcagc acctgagttt                                  | 20  |
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| <211>          | 20  |     |
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| <223>          | Description of Artificial Sequence: Synthetic DNA |     |
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| <400>          | 19  |     |
| gttgcg         | ggcca tatctaccag                                  | 20  |
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| <400>2         | 20  |     |
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| Luuarr         | attt teageeteta                                   | 20  |

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Val Pro Ser Glu Ser Val Thr Arg Lys Ser Gln Arg Arg Lys Ala Thr
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Ser Pro Gly Glu Ser Arg Glu Ser Ser Lys Asp Arg Leu Leu Ile Leu
Pro Ser Met Gly Glu Ser Tyr Thr Glu Tyr Val Asp Ser Tyr Leu Asn
Leu Glu Leu Leu Glu Arg Gly Glu Arg Glu Thr Pro Ile Phe Leu Glu
Ser Leu Thr Arg Gln Leu Thr Gln Lys Ile Tyr Glu Leu Ile Lys Thr
Lys Ser Leu Thr Ala Asp Ala Leu Gln Gln Ile Ser Asp Lys Tyr Asp
            100
Gly Val Val Ala Glu Asn Lys Leu Leu Phe Leu Gln Arg Gln Tyr Tyr
                            120
Val Asp Asp Glu Gly Asn Val Arg Asp Gly Arg Asn Asn Asp Lys Ile
    130
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Tyr Cys Glu Pro Lys His Val Tyr Asp Met Val Met Ala Thr His Leu 150 155 Met Asn Lys His Leu Arg Gly Lys Thr Leu His Ser Phe Leu Phe Ser 170 His Phe Ala Asn Ile Ser His Ala Ile Ile Asp Trp Val Gln Gln Phe Cys Ser Lys Cys Asn Lys Lys Gly Lys Ile Lys Pro Leu Lys Glu Tyr 200 Lys Arg Pro Asp Met Tyr Asp Lys Leu Leu Pro Met Glu Arg Ile His 215 Ile Glu Val Phe Glu Pro Phe Asn Gly Glu Ala Ile Glu Gly Lys Tyr 230 Ser Tyr Val Leu Leu Cys Arg Asp Tyr Arg Ser Ser Phe Met Trp Leu 250 Leu Pro Leu Lys Ser Thr Lys Phe Lys His Leu Ile Pro Val Val Ser Ser Leu Phe Leu Thr Phe Ala Arg Val Pro Ile Phe Val Thr Ser Ser 280 Thr Leu Asp Lys Asp Asp Leu Tyr Asp Ile Cys Glu Glu Ile Ala Ser 295 Lys Tyr Gly Leu Arg Ile Gly Leu Gly Leu Lys Ser Ser Ala Arg Phe 310 His Thr Gly Gly Ile Leu Cys Ile Gln Tyr Ala Leu Asn Ser Tyr Lys 330 Lys Glu Cys Leu Ala Asp Trp Gly Lys Cys Leu Arg Tyr Gly Pro Tyr Arg Phe Asn Arg Arg Asn Lys Arg Thr Lys Arg Lys Pro Val Gln Val Leu Leu Ser Glu Val Pro Gly His Asn Ala Lys Phe Glu Thr Lys 375 Arg Glu Arg Val Ile Glu Asn Thr Tyr Ser Arg Asn Met Phe Lys Met Ala Gly Gly Lys Gly Leu Ile Tyr Leu Glu Asp Val Asn Thr Phe Ala 410 Leu Ala Asn Glu Ala Asp Asn Ser Cys Asn Asn Asn Gly Ile Leu His 420 Asn Asn Asn Ile Gly Asn Asp Asn Phe Glu Glu Val Gln Lys Gln 440 445

Phe Asp Leu Thr Glu Lys Asn Tyr Ile Asp Glu Tyr Asp Asp Leu Ala 450 455 460

His Asp Ser Ser Glu Gly Glu Phe Glu Pro Asn Thr Leu Thr Pro Glu 465 470 475 480

Glu Lys Pro Pro His Asn Val Asp Glu Asp Arg Ile Glu Ser Thr Gly 485 490 495

Val Ala Ala Pro Met Gln Gly Thr Glu Glu Pro Glu Lys Gly Asp Gln
500 505 510

Lys Glu Ser Asp Gly Ala Ser Gln Val Asp Gln Ser Val Glu Ile Thr 515 520 525

Arg Pro Glu Thr Ser Tyr Tyr Gln Thr Leu Glu Ser Pro Ser Thr Lys 530 540

Arg Gln Lys Leu Asp Gln Gln Gly Asn Gly Asp Gln Thr Arg Asp Phe 545 550 555 560

Gly Thr Ser Met Glu Leu

<210> 26

<211> 309

<212> PRT

<213> Escherichia coli

<400> 26

Met Ala Arg Tyr Asp Leu Val Asp Arg Leu Asn Thr Thr Phe Arg Gln
1 5 10 15

Met Glu Gln Glu Leu Ala Ile Phe Ala Ala His Leu Glu Gln His Lys
20 25 30

Leu Leu Val Ala Arg Val Phe Ser Leu Pro Glu Val Lys Lys Glu Asp 35 40 45

Glu His Asn Pro Leu Asn Arg Ile Glu Val Lys Gln His Leu Gly Asn 50 55 60

Asp Ala Gln Ser Leu Ala Leu Arg His Phe Arg His Leu Phe Ile Gln 65 70 75 80

Gln Gln Ser Glu Asn Arg Ser Ser Lys Ala Ala Val Arg Leu Pro Gly 85 90 95

Val Leu Cys Tyr Gln Val Asp Asn Leu Ser Gln Ala Ala Leu Val Ser 100 105 110

His Ile Gln His Ile Asn Lys Leu Lys Thr Thr Phe Glu His Ile Val

Thr Val Glu Ser Glu Leu Pro Thr Ala Ala Arg Phe Glu Trp Val His 130 135 140 Arg His Leu Pro Gly Leu Ile Thr Leu Asn Ala Tyr Arg Thr Leu Thr 150 155 Val Leu His Asp Pro Ala Thr Leu Arg Phe Gly Trp Ala Asn Lys His 170 Ile Ile Lys Asn Leu His Arg Asp Glu Val Leu Ala Gln Leu Glu Lys Ser Leu Lys Ser Pro Arg Ser Val Ala Pro Trp Thr Arg Glu Glu Trp 200 Gln Arg Lys Leu Glu Arg Glu Tyr Gln Asp Ile Ala Ala Leu Pro Gln Asn Ala Lys Leu Lys Ile Lys Arg Pro Val Lys Val Gln Pro Ile Ala Arg Val Trp Tyr Lys Gly Asp Gln Lys Gln Val Gln His Ala Cys Pro 250 Thr Pro Leu Ile Ala Leu Ile Asn Arg Asp Asn Gly Ala Gly Val Pro 260 Asp Val Gly Glu Leu Leu Asn Tyr Asp Ala Asp Asn Val Gln His Arg Tyr Lys Pro Gln Ala Gln Pro Leu Arg Leu Ile Ile Pro Arg Leu His 295 300 290 Leu Tyr Val Ala Asp <210> 27 <211> 1698 <212> DNA <213> Saccharomyces cerevisiae <220> <221> CDS <222> (1)..(1698) <400> 27 atg acg aaa ccg cgt tac aat gac gtg ttg ttt gat gat gat gac tcg 48 Met Thr Lys Pro Arg Tyr Asn Asp Val Leu Phe Asp Asp Asp Asp Ser 1 gta cca tca gaa tca gtt acg agg aaa tcg cag aga aga aag gca acg 96 Val Pro Ser Glu Ser Val Thr Arg Lys Ser Gln Arg Arg Lys Ala Thr 20 tca cct ggg gaa tca aga gag tcc tca aaa gat cgt cta ctg ata ctt Ser Pro Gly Glu Ser Arg Glu Ser Ser Lys Asp Arg Leu Leu Ile Leu 35

|                   |            |            |                   |                   | tca<br>Ser        |            |            |                   |                   |                   |            |            |                   |                   |                   | 192 |
|-------------------|------------|------------|-------------------|-------------------|-------------------|------------|------------|-------------------|-------------------|-------------------|------------|------------|-------------------|-------------------|-------------------|-----|
|                   | _          |            |                   | _                 | agg<br>Arg<br>70  |            | _          | _                 | -                 |                   |            |            |                   |                   | _                 | 240 |
|                   | _          |            | _                 |                   | cta<br>Leu        | _          | _          |                   |                   |                   | _          |            |                   |                   |                   | 288 |
|                   |            |            |                   | _                 | gat<br>Asp        | _          | _          |                   |                   |                   | _          | _          |                   |                   | _                 | 336 |
|                   | -          |            | -                 | _                 | aac<br>Asn        | _          | -          |                   |                   | _                 |            | _          | _                 |                   |                   | 384 |
|                   |            |            |                   |                   | aat<br>Asn        |            |            |                   |                   |                   |            |            |                   |                   |                   | 432 |
|                   |            |            |                   |                   | cat<br>His<br>150 |            |            |                   |                   |                   |            |            |                   |                   |                   | 480 |
| _                 |            | _          |                   |                   | agg<br>Arg        |            |            |                   |                   |                   |            |            |                   |                   |                   | 528 |
|                   |            |            |                   |                   | agt<br>Ser        |            |            |                   |                   |                   |            |            |                   |                   |                   | 576 |
|                   |            |            |                   |                   | aaa<br>Lys        |            |            |                   |                   |                   |            |            |                   |                   |                   | 624 |
|                   |            |            |                   |                   | tac<br>Tyr        |            |            |                   |                   |                   |            |            |                   |                   |                   | 672 |
| att<br>Ile<br>225 | gag<br>Glu | gta<br>Val | ttc<br>Phe        | gaa<br>Glu        | ccc<br>Pro<br>230 | ttc<br>Phe | aat<br>Asn | gga<br>Gly        | gaa<br>Glu        | gct<br>Ala<br>235 | att<br>Ile | gag<br>Glu | gga<br>Gly        | aaa<br>Lys        | tat<br>Tyr<br>240 | 720 |
| tct<br>Ser        | tat<br>Tyr | gtc<br>Val | ctt<br>Leu        | tta<br>Leu<br>245 | tgc<br>Cys        | cga<br>Arg | gac<br>Asp | tat<br>Tyr        | cgc<br>Arg<br>250 | tct<br>Ser        | agt<br>Ser | ttt<br>Phe | atg<br>Met        | tgg<br>Trp<br>255 | tta<br>Leu        | 768 |
| tta<br>Leu        | cca<br>Pro | ctt<br>Leu | aag<br>Lys<br>260 | agt<br>Ser        | acc<br>Thr        | aaa<br>Lys | ttc<br>Phe | aaa<br>Lys<br>265 | cat<br>His        | ctt<br>Leu        | atc<br>Ile | cca<br>Pro | gtt<br>Val<br>270 | gtt<br>Val        | tcc<br>Ser        | 816 |

|   |   |   |   |   | ttt<br>Phe        |   |   |   |   |   |   |   |   |   |   | 864  |
|---|---|---|---|---|-------------------|---|---|---|---|---|---|---|---|---|---|------|
|   |   | _ |   | _ | gat<br>Asp        |   |   | _ |   | _ | _ | _ |   | _ |   | 912  |
|   |   |   |   | _ | att<br>Ile<br>310 |   | _ |   | _ | _ | _ |   |   | _ |   | 960  |
|   |   |   |   |   | ctg<br>Leu        | _ |   | _ |   |   |   |   | ~ |   | _ | 1008 |
|   |   |   |   |   | gat<br>Asp        |   |   |   |   |   |   |   |   |   |   | 1056 |
| _ |   |   | _ |   | aga<br>Arg        |   | _ | - | _ |   | _ |   |   |   |   | 1104 |
| _ |   |   | _ | _ | gtt<br>Val        |   |   |   |   | _ | _ |   |   |   | _ | 1152 |
| _ | _ |   | _ |   | gaa<br>Glu<br>390 |   |   |   |   | _ |   | _ |   | _ | _ | 1200 |
|   |   |   |   |   | ctt<br>Leu        |   |   |   |   |   |   |   |   |   |   | 1248 |
|   | _ |   | _ |   | gat<br>Asp        |   | _ | _ |   |   |   |   |   |   |   | 1296 |
|   |   |   |   |   | aat<br>Asn        |   |   |   |   |   |   |   |   |   |   | 1344 |
|   |   |   |   |   | aaa<br>Lys        |   |   |   |   |   |   |   |   |   |   | 1392 |
|   |   |   |   |   | ggc<br>Gly<br>470 |   |   |   |   |   |   |   |   |   |   | 1440 |
|   |   |   |   |   | aat<br>Asn        |   |   |   |   |   |   |   |   |   |   | 1488 |

| vai  | gca<br>Ala   |  | Pro  |  |  |  |   | Glu                                |  |   |  |   | Gly  |   |  | 1536             |
|--|--|--|--|--|--|--|---|------------------------------------|--|---|--|---|--|---|--|------------------|
|  |  |  | 500  |  |  |  |   | 505                                |  |   |  |   | 510  |   |  |                  |
|  | gaa<br>Glu   |  |  |  |  |  |   |                                    |  |   |  |   |  |   |  | 1584             |
|  | cca<br>Pro<br>530  |  |  |  |  |  |   |                                    |  |   |  |   |  |   |  | 1632             |
|  | cag<br>Gln   |  |  |  |  |  |   |                                    |  |   |  |   |  |   |  | 1680             |
|  | aca<br>Thr   |  | _  | _  | _                                      |  |   |                                    |  |   |  |   |  |   |  | 1698             |
| <210> 28 <211> 927 <212> DNA <213> Escherichia coli                                  |  |  |  |  |  |  |   |                                    |  |   |  |   |  |   |  |                  |
| <22  | <220> <221> CDS <222> (1)(927)                             |  |  |  |  |  |   |                                    |  |   |  |   |  |   |  |                  |
| <40  | 0- 20  |  |  |  |  |  |   |                                    |  |   |  |   |  |   |  |                  |
|  |  |  | <b>+</b>   | an t   | ata                                    | ~+ n                                       | <b>~</b> 2~   | ~~~                                | a+ a                                   | 224   | 20+                                    | 200   |  | 999   | 929  | 40               |
| atg  | gcg<br>Ala   | cgt  |  | _  |  | _  | _   | _                                  |  |   |  |   |  | _   | _  | 48               |
| atg<br>Met<br>1<br>atg   | gcg  | cgt<br>Arg   | Tyr<br>gag   | Asp<br>5<br>ctg                                    | Leu<br>gct                             | Val<br>ata                                 | Asp   | Arg                                | Leu<br>10<br>gct                       | Asn<br>cat                                    | Thr                                    | Thr<br>gag  | Phe<br>caa                                   | Arg<br>15<br>cac                                    | Gln  | 48<br>96         |
| atg<br>Met<br>1<br>atg<br>Met  | gcg<br>Ala<br>gaa  | cgt<br>Arg<br>caa<br>Gln   | Tyr<br>gag<br>Glu<br>20<br>gcc                             | Asp<br>5<br>ctg<br>Leu<br>cgc                      | Leu<br>gct<br>Ala<br>gtg               | Val<br>ata<br>Ile                          | Asp<br>ttt<br>Phe<br>tct  | Arg gcc Ala 25 ttg                 | Leu<br>10<br>gct<br>Ala<br>ccg         | Asn<br>cat<br>His                             | Thr<br>ctt<br>Leu<br>gta               | Thr<br>gag<br>Glu<br>aaa                            | Phe<br>caa<br>Gln<br>30<br>aaa               | Arg<br>15<br>cac<br>His                             | Gln<br>aag<br>Lys<br>gat                                   |                  |
| atg<br>Met<br>1<br>atg<br>Met<br>cta<br>Leu  | gcg<br>Ala<br>gaa<br>Glu                                   | cgt<br>Arg<br>caa<br>Gln<br>gtt<br>Val<br>35                             | gag<br>Glu<br>20<br>gcc<br>Ala                             | Asp<br>5<br>ctg<br>Leu<br>cgc<br>Arg               | gct<br>Ala<br>gtg<br>Val               | Val<br>ata<br>Ile<br>ttc<br>Phe            | Asp<br>ttt<br>Phe<br>tct<br>Ser<br>40                             | gcc<br>Ala<br>25<br>ttg<br>Leu     | Leu<br>10<br>gct<br>Ala<br>ccg<br>Pro  | Asn<br>cat<br>His<br>gag<br>Glu<br>aaa        | Thr<br>ctt<br>Leu<br>gta<br>Val        | Thr gag Glu aaa Lys 45 cat                          | Phe caa Gln 30 aaa Lys                       | Arg<br>15<br>cac<br>His<br>gag<br>Glu               | Gln aag Lys gat Asp  | 96               |
| atg<br>Met<br>1<br>atg<br>Met<br>cta<br>Leu<br>gag<br>Glu                            | gcg<br>Ala<br>gaa<br>Glu<br>ttg<br>Leu<br>cat              | cgt<br>Arg<br>caa<br>Gln<br>gtt<br>Val<br>35<br>aat<br>Asn               | gag<br>Glu<br>20<br>gcc<br>Ala<br>ccg<br>Pro               | Asp<br>5<br>ctg<br>Leu<br>cgc<br>Arg<br>ctt<br>Leu | gct<br>Ala<br>gtg<br>Val<br>aat<br>Asn | Val ata Ile ttc Phe cgt Arg 55             | Asp<br>ttt<br>Phe<br>tct<br>Ser<br>40<br>att<br>Ile               | gcc Ala 25 ttg Leu gag Glu cat     | Leu 10 gct Ala ccg Pro gta Val         | Asn<br>cat<br>His<br>gag<br>Glu<br>aaa<br>Lys | Thr  ctt Leu  gta Val  caa Gln 60 cat  | Thr<br>gag<br>Glu<br>aaa<br>Lys<br>45<br>cat<br>His | Caa<br>Gln<br>30<br>aaa<br>Lys<br>Ctc<br>Leu | Arg<br>15<br>cac<br>His<br>gag<br>Glu<br>ggc<br>Gly | aag<br>Lys<br>gat<br>Asp<br>aac<br>Asn                     | 96<br>144        |
| atg<br>Met<br>1<br>atg<br>Met<br>cta<br>Leu<br>gag<br>Glu<br>gac<br>Asp<br>65<br>caa | gcg<br>Ala<br>gaa<br>Glu<br>ttg<br>Leu<br>cat<br>His<br>50 | cgt<br>Arg<br>caa<br>Gln<br>gtt<br>Val<br>35<br>aat<br>Asn<br>cag<br>Gln | gag<br>Glu<br>20<br>gcc<br>Ala<br>ccg<br>Pro<br>tcg<br>ser | Asp 5 ctg Leu cgc Arg ctt Leu ctg Leu aat          | gct Ala gtg Val aat Asn gcg Ala 70 cgc | Val ata Ile ttc Phe cgt Arg 55 ttg Leu agc | Asp<br>ttt<br>Phe<br>tct<br>Ser<br>40<br>att<br>Ile<br>cgt<br>Arg | gcc Ala 25 ttg Leu gag Glu cat His | Leu 10 gct Ala ccg Pro gta Val ttc Phe | Asn cat His gag Glu aaa Lys cgc Arg 75        | Thr ctt Leu gta Val caa Gln 60 cat His | Thr gag Glu aaa Lys 45 cat His tta Leu cgt          | Caa Gln 30 aaa Lys Ctc Leu ttt Phe Ctg       | Arg 15 cac His gag Glu ggc Gly att Ile cct          | aag<br>Lys<br>gat<br>Asp<br>aac<br>Asn<br>caa<br>Gln<br>80 | 96<br>144<br>192 |

|            |            |            |            |            |            |            |            |            |            |            |            | gag<br>Glu<br>125 |            |            |            | 384 |
|------------|------------|------------|------------|------------|------------|------------|------------|------------|------------|------------|------------|-------------------|------------|------------|------------|-----|
|            |            |            |            |            |            |            |            |            |            |            |            | gaa<br>Glu        |            |            |            | 432 |
|            |            |            |            |            |            |            |            |            |            |            |            | cgc<br>Arg        |            |            |            | 480 |
| -          | _          |            | _          |            | _          |            |            | _          |            |            |            | gct<br>Ala        |            |            |            | 528 |
| Ile        | Ile        | Lys        | Asn<br>180 | Leu        | His        | Arg        | Asp        | Glu<br>185 | Val        | Leu        | Āla        | cag<br>Gln        | Leu<br>190 | Glu        | Lys        | 576 |
| Ser        | Leu        | Lys<br>195 | Ser        | Pro        | Arg        | Ser        | Val<br>200 | Ala        | Pro        | Trp        | Thr        | cgc<br>Arg<br>205 | Glu        | Glu        | Trp        | 624 |
| Gln        | Arg<br>210 | Lys        | Leu        | Glu        | Arg        | Glu<br>215 | Tyr        | Gln        | Asp        | Ile        | Ala<br>220 | gcc<br>Ala        | Leu        | Pro        | Gln        | 672 |
| Asn<br>225 | Ala        | Lys        | Leu        | Lys        | Ile<br>230 | Lys        | Arg        | Pro        | Val        | Lys<br>235 | Val        | cag<br>Gln        | Pro        | Ile        | Ala<br>240 | 720 |
| Arg        | Val        | Trp        | Tyr        | Lys<br>245 | Ğly        | Āsp        | Gln        | Lys        | Gln<br>250 | Val        | Gln        | cac<br>His        | Āla        | Cys<br>255 | Pro        | 768 |
| Thr        | Pro        | Leu        | Ile<br>260 | Ala        | Leu        | Ile        | Asn        | Arg<br>265 | Asp        | Asn        | Gly        | gcg<br>Ala        | Gly<br>270 | Val        | Pro        | 816 |
| Asp        | Val        | Gly<br>275 | Glu        | Leu        | Leu        | Asn        | Tyr<br>280 | Asp        | Ala        | Asp        | Asn        | gtg<br>Val<br>285 | Gln        | His        | Arg        | 864 |
|            |            |            |            |            |            |            |            |            |            |            |            | cca<br>Pro        |            |            |            | 912 |
|            |            | gtt<br>Val |            |            |            |            |            |            |            |            |            |                   |            |            |            | 927 |